

Q 11 primers with [<sup>32</sup>P]-ATP. The amplification products were separated in a denaturing polyacrylamide gel electrophoresis, and analyzed by phosphorimaging. - -

IN THE CLAIMS:

Kindly cancel claims 1-50, and add new claims 51-131 set forth below:

Sub B1 51. A method for producing a nuclear transfer unit having genomic DNA of one mammalian species and mitochondria of a different mammalian species, comprising:

Q 12 (i) removing the genomic DNA from a mammalian oocyte;

(ii) inserting a differentiated mammalian donor cell, or the nucleus of said cell, into the oocyte under conditions suitable for the formation of a nuclear transfer unit so that a nuclear transfer unit is formed, wherein said oocyte and said differentiated cell are from different mammalian species;

(iii) activating the resultant nuclear transfer unit; and

(iv) culturing the activated nuclear transfer unit to produce a multicellular structure.

52. The method of claim 51, wherein step (ii) further comprises fusing the differentiated cell and the oocyte.

53. The method of claim 52, wherein fusion is effected by electrofusion.

54. The method of claim 51, wherein the step of activating the nuclear transfer unit comprises exposing said nuclear transfer unit to an ionophore.

55. The method of claim 51, wherein said differentiated donor cell is a non-embryonic cell.

56. The method of claim 51 wherein the differentiated donor cell is a germ cell.

57. The method of Claim 51, wherein the differentiated donor cell is a somatic cell.

58. The method of claim 51, wherein the differentiated donor cell is selected from the group consisting of epithelial cells, neural cells, epidermal cells, keratinocytes, hematopoietic cells, melanocytes, chondrocytes, B lymphocytes, T lymphocytes, erythrocytes, macrophages, monocytes, mononuclear cells, fibroblasts, and muscle cells.

59. The method of claim 51, wherein the differentiated donor cell is from an organ selected from the group consisting of skin, lung, pancreas, liver, stomach, intestine, heart, reproductive organs, bladder, kidney, urethra, and other urinary organs.

60. The method of claim 51 wherein the differentiated donor cell is a fibroblast.

61. The method of claim 51 wherein the differentiated donor cell is a human cell.

62. The method of claim 61 wherein the differentiated donor cell is a human epithelial cell or a human keratinocyte.

63. The method of claim 51 wherein the differentiated donor cell is from an ungulate.

64. The method of claim 51, wherein the oocyte is from a mammal selected from the group consisting of sheep, bovines, ovines, pigs, horses, rabbits, goats, guinea pigs, mice, hamsters, rats, and primates.

B2  
concur  
65. The method of claim 64, wherein the oocyte is from a primate.

66. The method of claim 51, wherein the oocyte is from an ungulate.

A12  
67. The method of claim 66, wherein the oocyte is from an ungulate selected from the group consisting of bovines, ovines, porcines, equines, caprines, and buffalo.

B  
68. The method of claim 67, wherein the oocyte is a bovine oocyte.

Sub  
633  
69. The method of claim 51, wherein the differentiated donor cell is a human cell and the oocyte is a bovine oocyte.

70. The method of claim 51, comprising culturing said activated nuclear transfer unit on a feeder layer of fibroblast cells to produce a multicellular structure.

71. The method of claim 51, further comprising isolating an embryonic cell from the multicellular structure produced by the cultured nuclear transfer unit.

72. The method of claim 71, wherein the embryonic cell is isolated from a multicellular structure of about 2 to 400 cells.

73. The method of claim 51, further comprising culturing said activated nuclear transfer unit to produce a blastocyst.

74. The method of claim 73, further comprising isolating embryonic cells from the blastocyst.

75. The method of claim 74, further comprising culturing an embryonic cell isolated from the blastocyst, and producing a cell line from said embryonic cell.

Q<sup>12</sup> 76. The method of claim 51, wherein the genome of the differentiated donor cell is genetically altered by addition, modification, substitution, or deletion of one or more genes.

77. The method of claim 76, wherein the genome of the donor cell is genetically altered by a method comprising homologous recombination.

78. The method of claim 76, wherein the genome of the differentiated donor cell is genetically altered by addition, modification, substitution, or deletion of one or more genes that encode an enzyme, a growth factor, or a cytokine.

79. An isolated embryonic cell produced by the method of claim 71, which cell is not itself an embryo.

80. The isolated embryonic cell of claim 79, which cell has non-bovine genomic DNA and bovine mitochondria.

81. The isolated embryonic cell of claim 79, which cell has human genomic DNA and mitochondria of a non-human mammal.

82. The isolated embryonic cell of claim 79, which cell has human genomic DNA and bovine mitochondria.

B4  
Q12  
83. An isolated embryonic cell which is not itself an embryo, which cell has genomic DNA of one species of mammal and mitochondria of a different species of mammal.

84. The isolated embryonic cell of claim 83, which cell has human genomic DNA and non-human mitochondria.

85. The isolated embryonic cell of claim 83, which cell has human genomic DNA and bovine mitochondria.

86. A method for producing a nuclear transfer unit having genetically altered genomic DNA of one mammalian species and mitochondria of a different mammalian species, comprising:

- (i) obtaining a differentiated mammalian donor cell, the genome of which is genetically altered by addition, modification, substitution, or deletion of one or more genes;
- (ii) removing the genomic DNA from a mammalian oocyte;
- (iii) inserting the genetically altered donor cell, or the nucleus of said cell, into the oocyte under conditions suitable for the formation of a nuclear transfer unit so that a nuclear

transfer unit is formed, wherein said oocyte and said differentiated donor cell are from different mammalian species;

(iv) activating the resultant nuclear transfer unit; and

*But same*  
(v) culturing the activated nuclear transfer unit to produce a multicellular structure.

87. The method of claim 86, wherein the genome of the donor cell is genetically altered by a method comprising homologous recombination.

*Q12*  
88. The method of claim 86, wherein the genome of the donor cell is genetically altered by addition, modification, substitution, or deletion of one or more genes that encode an enzyme, a growth factor, or a cytokine.

89. The method of claim 86, wherein fusion is effected by electrofusion.

90. The method of claim 86, wherein the step of activating the nuclear transfer unit comprises exposing said nuclear transfer unit to an ionophore.

91. The method of claim 86, wherein the differentiated donor cell is a non-embryonic cell.

92. The method of claim 86 wherein the differentiated donor cell is a germ cell.

93. The method of Claim 86, wherein the differentiated donor cell is a somatic cell.

94. The method of claim 86, wherein the differentiated donor cell is selected from the group consisting of epithelial cells, neural cells, epidermal cells, keratinocytes, hematopoietic cells, melanocytes, chondrocytes, B lymphocytes, T lymphocytes, erythrocytes, macrophages, monocytes, mononuclear cells, fibroblasts, and muscle cells.

95. The method of claim 86, wherein the differentiated donor cell is from an organ selected from the group consisting of skin, lung, pancreas, liver, stomach, intestine, heart, reproductive organs, bladder, kidney, urethra, and other urinary organs.

Q12 96. The method of claim 86 wherein the differentiated donor cell is a fibroblast.

97. The method of claim 86 wherein the differentiated donor cell is a human cell.

98. The method of claim 86 wherein the differentiated donor cell is a human epithelial cell or a human keratinocyte.

99. The method of claim 86 wherein the differentiated donor cell is from an ungulate.

100. The method of claim 86, wherein the oocyte is from a mammal selected from the group consisting of sheep, bovines, ovines, pigs, horses, rabbits, goats, guinea pigs, mice, hamsters, rats, and primates

101. The method of claim 100, wherein the oocyte is from a primate.

35 June  
102. The method of claim 86, wherein the oocyte is from an ungulate.

103. The method of claim 102, wherein the oocyte is from an ungulate selected from the group consisting of bovines, ovines, porcines, equines, caprines, and buffalo.

B  
104. The method of claim 103, wherein the oocyte is a bovine oocyte.

gubio  
a 12  
105. The method of claim 86, wherein the differentiated donor cell is a human cell and the oocyte is a bovine oocyte.

106. The method of claim 86, comprising culturing said activated nuclear transfer unit on a feeder layer of fibroblast cells to produce a multicellular structure.

107. The method of claim 86, further comprising isolating an embryonic cell from the multicellular structure produced by the cultured nuclear transfer unit.

108. The method of claim 107, wherein the embryonic cell is isolated from a multicellular structure of about 2 to 400 cells.

109. The method of claim 86, further comprising culturing said activated nuclear transfer unit to produce a blastocyst.

110. The method of claim 109, further comprising isolating embryonic cells from the blastocyst.



111. The method of claim 110, further comprising culturing an embryonic cell having a genetically altered genome isolated from the blastocyst, and producing a cell line from said embryonic cell.

112. An isolated cell having genetically altered genomic DNA produced by the method of claim 107, which cell is not itself an embryo.

sub B7  
112  
113. The isolated cell of claim 112, which cell has genetically altered, non-bovine genomic DNA and bovine mitochondria.

114. The isolated cell of claim 112, which cell has genetically altered, human genomic DNA and mitochondria of a non-human mammal.

115. The isolated cell of claim 112, which cell has genetically altered, human genomic DNA and bovine mitochondria.

116. The isolated cell of claim 112, the genome of which is genetically altered by addition, modification, substitution, or deletion of one or more genes that encode an enzyme, a growth factor, or a cytokine.

sub B8  
117. An isolated embryonic cell which is not itself an embryo, which cell has genetically altered genomic DNA of one species of mammal and mitochondria of a different species of mammal.

118. The isolated embryonic cell of claim 117, which cell has genetically altered, human genomic DNA and non-human mitochondria.

B8  
com  
119. The isolated embryonic cell of claim 117, which cell has genetically altered, human genomic DNA and bovine mitochondria.

120. The isolated embryonic cell of claim 117, the genome of which is genetically altered by addition, modification, substitution, or deletion of one or more genes that encode an enzyme, a growth factor, or a cytokine.

Q12  
121. A cell of the cell line produced by the method of claim 75.

Sub  
B9  
122. The cell of claim 121, which cell has non-bovine genomic DNA and bovine mitochondria.

123. The cell of claim 121, which cell has human genomic DNA and mitochondria of a non-human mammal.

124. The cell of claim 121, which cell has human genomic DNA and bovine mitochondria.

125. The method of claim 75, further comprising genetically altering the genomic DNA of a cell of said cell line by adding, modifying, substituting, or deleting one or more genes.

126. The method of claim 125, wherein the genome of said cell is genetically altered by addition, modification, substitution, or deletion of one or more genes that encode an enzyme, a growth factor, or a cytokine.

127. The method of claim 125, wherein the genome of said cell is genetically altered by a method comprising homologous recombination.

128. A cell having genetically altered genomic DNA produced by the method of claim 125.

Q<sup>12</sup>  
Sub 120  
129. The cell of claim 128, which cell has genetically altered, non-bovine genomic DNA and bovine mitochondria.

Rule  
1.126  
130. The cell of claim 128, which cell has genetically altered, human genomic DNA and mitochondria of a non-human mammal.

1  
131. The cell of claim 128, which cell has genetically altered, human genomic DNA and bovine mitochondria.

2  
132. The cell of claim 128, the genome of which is genetically altered by addition, modification, substitution, or deletion of one or more genes that encode an enzyme, a growth factor, or a cytokine.

---